EFFECTS OF GROWTH REGULATORS ON ROOT TIP CELLS OF ONION

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Abstract

Cell size and mitotic index were decreased with increasing concentrations of growth regulators such as GA_3 , uniconazole, chlorocholine chloride (CCC) and 2,3,6-TBA while nuclear volume, interphase chromosome volume and abnormalities were found to increase during division of onion root tip cells. Formation of chromosome fragments, bridges, laggards, single and multiple chromatid bridges, irregular distribution and unequal separation of chromosomes were visualized as the main effects due to the application of these growth regulators.

Radiomimetic action of growth regulators such as hormones have been demonstrated by several workers (Prasad and Das 1977, Bebars 1987, Barrett and Nell 1992). Some of the growth regulators are used as growth promoters while others as growth retardants and herbicides. Uniconazole and CCC are plant growth retardants belonging to a group of triazoles. There is a little retarding effect of 2,3,6-TBA while GA₃ stimulates growth. Growth retardants are diverse in nature and have the common physiological effect of reducing stem growth by inhibiting cell division of the subapical meristem. Henry (1985) reported that uniconazole reduced plant height by inhibiting GA biosynthesis. Coolbaugh and Hamilton (1976) showed that ancymidol (a growth retardant) also inhibited gibberellin biosynthesis. In this context it is necessary to study the comparative effects of different growth regulators under identical condition. Therefore, the aim of this work was to study the comparative effects of four growth regulators at different concentrations on cell division using onion root tip cell.

A local variety of onion called "Taherpuri" was used as experimental materials and four growth regulators, namely Gibberellic acid-GA₃ (growth promoter), uniconazole (growth retardant), chlorocholine chloride (CCC, growth retardant) and 2,3,6-trichlorobenzoic acid-2,3,6 (TBA, growth retardant and herbicidal) were used. Each growth substance was applied at five different concentrations such as 100, 150, 200, 250 and 300 ppm. During the treatment the bulbs of onion were placed on the mouth of each specimen tubes until the roots grew up to 1.0 to 1.5 cm in length. One tube was filled with water as control. Root tips were collected and fixed in 1:3 acetic acid-alcohol for 48 hours and stored in 70% ethanol in a refrigerator. Chromosomes were stained with 0.5% haematoxylin following the method of Haque *et al.* (1976). The nuclear volume (NV) was calculated using the formula as suggested by Nayar *et al.* (1971). The recorded data were analyzed statistically following MSTAT-C package program.

The comparative effects of four growth regulators on cell (differentiated) size, interphase chromosome volume (ICV), mitotic index and chromosome abnormalities are shown in Table 1 and Figs. 1-5.

The mean data revealed that highest dose (300 ppm) of uniconazole produced lowest cell size and mitotic index. while highest value of ICV and chromosomal abnormalities were observed compared to control and other growth regulators used (Table 1).

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Treat- ments	Doses (ppm)	Cell (differentiated) size (µm ²)	Mitotic index (%)	Interphase chromosome volume (μm ³) ICV=NV/2n	Chromosomal abnormalities (%)
Water	-	75.23a	38.91a	0.64k	0.15i
GA ₃	100	67.00fg	35.12e	2.10h-j	0.75f-i
	150	66.89fg	35.00e	2.43g-j	0.98f-i
	200	66.70fg	35.00e	3.19e-h	1.02f-i
	250	66.50fg	33.92e	3.53d-g	1.32e-h
	300	58.89ij	33.00e	3.58d-g	1.60ef
	Mean	65.20	34.40	2.96	1.13
Unicona-	100	62.18h	22.12f	4.04c-f	1.54e-g
zole	150	58.91ij	20.17g	4.60b-d	2.63cd
	200	55.25k	19.80i	5.75ab	3.39c
	250	50.12m	18.82k	5.82ab	4.46b
	300	42.500	15.051	6.23a	6.26a
	Mean	53.79	19.19	5.28	3.65
CCC	100	65.23g	25.25f	3.58d-g	1.01f-i
	150	60.37i	24.75f	3.67d-g	1.36e-h
	200	58.11j	22.90h	3.85d-f	2.09de
	250	52.721	22.00j	4.19с-е	3.10c
	300	47.78n	20.10k	5.21a-c	4.91b
	Mean	56.84	23.00	4.10	2.49
2,3,6-TBA	100	71.93b	27.12b	1.07jk	0.20i
	150	70.85c	26.70bc	1.24jk	0.25i
	200	68.92d	25.50b-d	1.29jk	0.41hi
	250	67.99e	24.12de	1.80i-k	0.45hi
	300	67.61f	24.00de	2.37f-i	0.50g-i
	Mean	69.46	25.48	1.55	0.36
CV (%)		1.58	1.98	10.22	9.36

Table 1. Effects of different growth regulators along with control on cell (differentiated) size, mitotic index, interphase chromosome volume and chromosomal abnormalities in the root tip cells of onion.

Means followed by the same letter do not statistically differ at 5% level tested by DMRT.

The cell size showed highly significant and positive correlation with mitotic index but highly significant and negative correlation with ICV (Table 2). It has non-significant negative correlation with chromosomal abnormalities. ICV showed highly significant and negative correlation with mitotic index but significant and positive correlation with chromosomal abnormalities. On the other hand mitotic index showed highly significant and negative correlation with chromosomal abnormalities (Table 2).

Characters	Cell size (µm ²)	ICV (µm ³)	Mitotic index (%)	Chromosomal abnormalities (%)
Cell size (µm ²)	-	-0.784**	0.706**	-0.446
ICV (µm ³)		-	-0.810**	0.682*
Mitotic index (%)			-	-0.895**
Chromosomal abnormalities (%)				-

 Table 2. Correlation coefficient (r) of different characters from the root tip cells of onion observed following application of different growth regulators.

* and ** indicate significance at 5 and 1% level, respectively.

It was found that all the growth regulators made the division of meristametic cells very slow and reduced length and breath of the differentiated cells. The role of gibberellins in cell elongation is still obscure. Sometimes it may cause cell elongation by the induction of enzyme that weakens the cell walls (MacLeod and Miller, 1962). Paleg (1965) reported that gibberellic acid increased number of dividing cells and cell size in internodes and that caused stem elongation but retarded rooting. Bebars (1987) noted that 0.05% chlormequat stimulated the mitotic index in root tip cells of onion whereas it was reduced at 0.025 and 0.100% concentrations. These findings supported the present findings and also indicated the inhibiting nature of plant growth regulators to some extent.



Figs. 1-5. Photomicrographs showing chromosomal abnormalities in root tip cells of onion treated by different doses of growth regulators. 1. Metaphase chromosomes with chromosome fragment and a small ring chromosome (× 750), 2. Anaphase (late) with chromosome fragment (× 850), 3. Anaphase (late) with chromatid bridge (× 750), 4. Anaphase (late) with chromatid bridge (broken) and fragment (× 750), 5. Telophase with chromatid bridge and fragment (× 750).

Prasad and Das (1977) reported chromosome breakage, bridge formation, C-mitosis, micronuclei formation, stickiness and chromosome condensation in root tip cells of *Vicia faba* due to treatment with GA, 2,4-D and IBA. The present findings revealed that the chromosomal abnormalities were found to increase with increased doses of all growth regulators. Similar results were reported by Wuu and Grant (1967) in the root tip cells of the C₁ generation in barley seeds treated with B-995. It is evident that growth regulators have different kinds of impact on cells even at low doses. Therefore, the recommended dose for each growth regulators must be applied.

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